



SEQUENCES

<110> NATIONAL INSTITUTE FOR AGRICULTURAL RESEARCH (INRA)

<120> PROCEDURE FOR THE PREPARATION OF 1,3-PROPANEDIOL STARTING FROM A RECOMBINANT MICRO-ORGANISM, IN THE ABSENCE OF COENZYME B12 OR ONE OF ITS PRECURSORS.

<130> CHEP004US

<140> PCT/FR00/01981

<141> 2000-07-07

<160> 10

<170> Patent IN Ver. 2.1.

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| | | | | | | | | | | | | | | | | | |
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Val Ala Val Lys Lys Ile Val Phe Asp Glu Asn Lys Ile Thr Pro Ser
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Val Asp Asn Leu Ala Arg Glu Gly Ala Leu Val Tyr Cys Arg Glu Val
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Pro Ser Ser Ile Asn Val Tyr Phe Gly Ser Leu Thr Gly Ala Thr Pro
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Asp Gly Arg Lys Ser Gly Gln Pro Leu Ala Asp Gly Val Ser Pro Ser
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690 695 700

Phe His Pro Ser Ala Leu Lys Gly Asp Asn Gly Leu Met Asn Leu Ser
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Ser Leu Ile Arg Ser Tyr Phe Asp Gln Lys Gly Phe His Val Gln Phe
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Asn Val Ile Asp Lys Lys Ile Leu Leu Ala Ala Gln Lys Asn Pro Glu
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Lys Tyr Gln Asp Leu Ile Val Arg Val Ala Gly Tyr Ser Ala Gln Phe
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His Val Met
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<213> Clostridium butyricum

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| Met | Ser | Lys | Glu | Ile | Lys | Gly | Val | Leu | Phe | Asn | Ile | Gln | Lys | Phe | Ser | 1 | 5 | 10 | 15 |
| Leu | His | Asp | Gly | Pro | Gly | Ile | Arg | Thr | Ile | Val | Phe | Phe | Lys | Gly | Cys | 20 | 25 | 30 | |
| Ser | Met | Ser | Cys | Leu | Trp | Cys | Ser | Asn | Pro | Glu | Ser | Gln | Asp | Ile | Lys | 35 | 40 | 45 | |
| Pro | Gln | Val | Met | Phe | Asn | Lys | Asn | Leu | Cys | Thr | Lys | Cys | Gly | Arg | Cys | 50 | 55 | 60 | |
| Lys | Ser | Gln | Cys | Lys | Ser | Ala | Gly | Ile | Asp | Met | Asn | Ser | Glu | Tyr | Arg | 65 | 70 | 75 | 80 |
| Ile | Asp | Lys | Ser | Lys | Cys | Thr | Glu | Cys | Thr | Lys | Cys | Val | Asp | Asn | Cys | 85 | 90 | 95 | |
| Leu | Ser | Gly | Ala | Leu | Val | Ile | Glu | Gly | Arg | Asn | Tyr | Ser | Val | Glu | Asp | 100 | 105 | 110 | |
| Val | Ile | Lys | Glu | Leu | Lys | Lys | Asp | Ser | Val | Gln | Tyr | Arg | Arg | Ser | Asn | 115 | 120 | 125 | |
| Gly | Gly | Ile | Thr | Leu | Ser | Gly | Gly | Glu | Val | Leu | Leu | Gln | Pro | Asp | Phe | 130 | 135 | 140 | |
| Ala | Val | Glu | Leu | Leu | Lys | Glu | Cys | Lys | Ser | Tyr | Gly | Trp | His | Thr | Ala | 145 | 150 | 155 | 160 |
| Ile | Glu | Thr | Ala | Met | Tyr | Val | Asn | Ser | Glu | Ser | Val | Lys | Lys | Val | Ile | 165 | 170 | 175 | |
| Pro | Tyr | Ile | Asp | Leu | Ala | Met | Ile | Asp | Ile | Lys | Ser | Met | Asn | Asp | Glu | 180 | 185 | 190 | |
| Ile | His | Arg | Lys | Phe | Thr | Gly | Val | Ser | Asn | Glu | Ile | Ile | Leu | Gln | Asn | 195 | 200 | 205 | |
| Ile | Lys | Leu | Ser | Asp | Glu | Leu | Ala | Lys | Glu | Ile | Ile | Ile | Arg | Ile | Pro | 210 | 215 | 220 | |
| Val | Ile | Glu | Gly | Phe | Asn | Ala | Asp | Leu | Gln | Ser | Ile | Gly | Ala | Ile | Ala | 225 | 230 | 235 | 240 |
| Gln | Phe | Ser | Lys | Ser | Leu | Thr | Asn | Leu | Lys | Arg | Ile | Asp | Leu | Leu | Pro | 245 | 250 | 255 | |
| Tyr | His | Asn | Tyr | Gly | Glu | Asn | Lys | Tyr | Gln | Ala | Ile | Gly | Arg | Glu | Tyr | 260 | 265 | 270 | |
| Ser | Leu | Lys | Glu | Leu | Lys | Ser | Pro | Ser | Lys | Asp | Lys | Met | Glu | Arg | Leu | 275 | 280 | 285 | |

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<212> PRT

<213> Clostridium butyricum

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Ala Arg Glu Asn Met Ala Tyr Ala Ser Leu Leu Ala Gly Met Ala Phe
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Glu His Met Ala Glu Leu Ala Leu Leu Asp Gly Asn Ala Phe Ser Asn
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<120> Procédé de préparation du 1,3-propanediol à partir d'un microorganisme recombinant, en l'absence de coenzyme B12 ou de l'un de ses précurseurs.

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Glu Leu Asp Arg Leu Asn Lys Arg Thr Gly Asp Ala Phe Gln Ile Ser

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Glu Glu Ser Lys Glu Lys Leu Lys Asp Val Phe Glu Tyr Trp Asn Gly

115

120

125

Lys Thr Thr Ser Glu Leu Ala Thr Ser Tyr Met Thr Glu Glu Thr Arg

130

135

140

Glu Ala Val Asn Cys Glu Val Phe Thr Val Gly Asn Tyr Tyr Tyr Asn

145

150

155

160

Gly Val Gly His Val Ser Val Asp Tyr Gly Lys Val Leu Arg Val Gly

165

170

175

Phe Asn Gly Ile Ile Asn Glu Ala Lys Glu Gln Leu Glu Lys Asn Arg

180

185

190

Ser Ile Asp Pro Asp Phe Ile Lys Lys Glu Lys Phe Leu Asn Ser Val

195

200

205

Ile Ile Ser Cys Glu Ala Ala Ile Thr Tyr Val Asn Arg Tyr Ala Lys

210

215

220

Lys Ala Lys Glu Ile Ala Asp Asn Thr Ser Asp Ala Lys Arg Lys Ala

225

230

235

240

Glu Leu Asn Glu Ile Ala Lys Ile Cys Ser Lys Val Ser Gly Glu Gly

245

250

255

Ala Lys Ser Phe Tyr Glu Ala Cys Gln Leu Phe Trp Phe Ile His Ala

260

265

270

Ile Ile Asn Ile Glu Ser Asn Gly His Ser Ile Ser Pro Ala Arg Phe

275

280

285

Asp Gln Tyr Met Tyr Pro Tyr Tyr Glu Asn Asp Lys Asn Ile Thr Asp

290

295

300

Lys Phe Ala Gln Glu Leu Ile Asp Cys Ile Trp Ile Lys Leu Asn Asp

305

310

315

320

Ile Asn Lys Val Arg Asp Glu Ile Ser Thr Lys His Phe Gly Gly Tyr

325

330

335

Pro Met Tyr Gln Lys Leu Ile Val Gly Gly Gln Asn Ser Glu Gly Lys

340

345

350

Asp Ala Thr Asn Lys Val Ser Tyr Met Ala Leu Glu Ala Ala Val His

355

360

365

Val Lys Leu Pro Gln Pro Ser Leu Ser Val Arg Ile Trp Asn Lys Thr

370

375

380

Pro Asp Glu Phe Leu Leu Arg Ala Ala Glu Leu Thr Arg Glu Gly Leu

385

390

395

400

Gly Leu Pro Ala Tyr Tyr Asn Asp Glu Val Ile Ile Pro Ala Leu Val

405

410

415

Ser Arg Gly Leu Thr Leu Glu Asp Ala Arg Asp Tyr Gly Ile Ile Gly

420

425

430

Cys Val Glu Pro Gln Lys Pro Gly Lys Thr Glu Gly Trp His Asp Ser

435

440

445

Ala Phe Phe Asn Leu Ala Arg Ile Val Glu Leu Thr Ile Asn Ser Gly

450

455

460

Phe Asp Lys Asn Lys Gln Ile Gly Pro Lys Thr Gln Asn Phe Glu Glu

465 470 475 480

Met Lys Ser Phe Asp Glu Phe Met Lys Ala Tyr Lys Ala Gln Met Glu

485 490 495

Tyr Phe Val Lys His Met Cys Cys Ala Asp Asn Cys Ile Asp Ile Ala

500 505 510

His Ala Glu Arg Ala Pro Leu Pro Phe Leu Ser Ser Met Val Asp Asn

515 520 525

Cys Ile Gly Lys Gly Lys Ser Leu Gln Asp Gly Gly Ala Glu Tyr Asn

530 535 540

Phe Ser Gly Pro Gln Gly Val Gly Val Ala Asn Ile Gly Asp Ser Leu

545 550 555 560

Val Ala Val Lys Lys Ile Val Phe Asp Glu Asn Lys Ile Thr Pro Ser

565 570 575

Glu Leu Lys Lys Thr Leu Asn Asn Asp Phe Lys Asn Ser Glu Glu Ile

580 585 590

Gln Ala Leu Leu Lys Asn Ala Pro Lys Phe Gly Asn Asp Ile Asp Glu

595 600 605

Val Asp Asn Leu Ala Arg Glu Gly Ala Leu Val Tyr Cys Arg Glu Val

610 615 620

Asn Lys Tyr Thr Asn Pro Arg Gly Gly Asn Phe Gln Pro Gly Leu Tyr

625 630 635 640

Pro Ser Ser Ile Asn Val Tyr Phe Gly Ser Leu Thr Gly Ala Thr Pro

645

650

655

Asp Gly Arg Lys Ser Gly Gln Pro Leu Ala Asp Gly Val Ser Pro Ser

660

665

670

Arg Gly Cys Asp Val Ser Gly Pro Thr Ala Ala Cys Asn Ser Val Ser

675

680

685

Lys Leu Asp His Phe Ile Ala Ser Asn Gly Thr Leu Phe Asn Gln Lys

690

695

700

Phe His Pro Ser Ala Leu Lys Gly Asp Asn Gly Leu Met Asn Leu Ser

705

710

715

720

Ser Leu Ile Arg Ser Tyr Phe Asp Gln Lys Gly Phe His Val Gln Phe

725

730

735

Asn Val Ile Asp Lys Lys Ile Leu Leu Ala Ala Gln Lys Asn Pro Glu

740

745

750

Lys Tyr Gln Asp Leu Ile Val Arg Val Ala Gly Tyr Ser Ala Gln Phe

755

760

765

Ile Ser Leu Asp Lys Ser Ile Gln Asn Asp Ile Ile Ala Arg Thr Glu

770

775

780

His Val Met

785

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<212> PRT

<213> Clostridium butyricum

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Leu His Asp Gly Pro Gly Ile Arg Thr Ile Val Phe Phe Lys Gly Cys

20

25

30

Ser Met Ser Cys Leu Trp Cys Ser Asn Pro Glu Ser Gln Asp Ile Lys

35

40

45

Pro Gln Val Met Phe Asn Lys Asn Leu Cys Thr Lys Cys Gly Arg Cys

50

55

60

Lys Ser Gln Cys Lys Ser Ala Gly Ile Asp Met Asn Ser Glu Tyr Arg

65

70

75

80

Ile Asp Lys Ser Lys Cys Thr Glu Cys Thr Lys Cys Val Asp Asn Cys

85

90

95

Leu Ser Gly Ala Leu Val Ile Glu Gly Arg Asn Tyr Ser Val Glu Asp

100

105

110

Val Ile Lys Glu Leu Lys Lys Asp Ser Val Gln Tyr Arg Arg Ser Asn

115

120

125

Gly Gly Ile Thr Leu Ser Gly Gly Glu Val Leu Leu Gln Pro Asp Phe

130

135

140

Ala Val Glu Leu Leu Lys Glu Cys Lys Ser Tyr Gly Trp His Thr Ala
145 150 155 160

Ile Glu Thr Ala Met Tyr Val Asn Ser Glu Ser Val Lys Lys Val Ile
165 170 175

Pro Tyr Ile Asp Leu Ala Met Ile Asp Ile Lys Ser Met Asn Asp Glu
180 185 190

Ile His Arg Lys Phe Thr Gly Val Ser Asn Glu Ile Ile Leu Gln Asn
195 200 205

Ile Lys Leu Ser Asp Glu Leu Ala Lys Glu Ile Ile Ile Arg Ile Pro
210 215 220

Val Ile Glu Gly Phe Asn Ala Asp Leu Gln Ser Ile Gly Ala Ile Ala
225 230 235 240

Gln Phe Ser Lys Ser Leu Thr Asn Leu Lys Arg Ile Asp Leu Leu Pro
245 250 255

Tyr His Asn Tyr Gly Glu Asn Lys Tyr Gln Ala Ile Gly Arg Glu Tyr
260 265 270

Ser Leu Lys Glu Leu Lys Ser Pro Ser Lys Asp Lys Met Glu Arg Leu
275 280 285

Lys Ala Leu Val Glu Ile Met Gly Ile Pro Cys Thr Ile Gly Ala Glu
290 295 300

<210> 8

<211> 385

<212> PRT

<213> Clostridium butyricum

<400> 8

Met Arg Met Tyr Asp Tyr Leu Val Pro Ser Val Asn Phe Met Gly Ala

1 5 10 15

Asn Ser Val Ser Val Val Gly Glu Arg Cys Lys Ile Leu Gly Gly Lys

20 25 30

Lys Ala Leu Ile Val Thr Asp Lys Phe Leu Lys Asp Met Glu Gly Gly

35 40 45

Ala Val Glu Leu Thr Val Lys Tyr Leu Lys Glu Ala Gly Leu Asp Val

50 55 60

Val Tyr Tyr Asp Gly Val Glu Pro Asn Pro Lys Asp Val Asn Val Ile

65 70 75 80

Glu Gly Leu Lys Ile Phe Lys Glu Glu Asn Cys Asp Met Ile Val Thr

85 90 95

Val Gly Gly Gly Ser Ser His Asp Cys Gly Lys Gly Ile Gly Ile Ala

100 105 110

Ala Thr His Glu Gly Asp Leu Tyr Asp Tyr Ala Gly Ile Glu Thr Leu

115 120 125

Val Asn Pro Leu Pro Pro Ile Val Ala Val Asn Thr Thr Ala Gly Thr

130 135 140

Ala Ser Glu Leu Thr Arg His Cys Val Leu Thr Asn Thr Lys Lys Lys

145 150 155 160

Ile Lys Phe Val Ile Val Ser Trp Arg Asn Leu Pro Leu Val Ser Ile

165 170 175

Asn Asp Pro Met Leu Met Val Lys Lys Pro Ala Gly Leu Thr Ala Ala

180

185

190

Thr Gly Met Asp Ala Leu Thr His Ala Ile Glu Ala Tyr Val Ser Lys

195

200

205

Asp Ala Asn Pro Val Thr Asp Ala Ser Ala Ile Gln Ala Ile Lys Leu

210

215

220

Ile Ser Gln Asn Leu Arg Gln Ala Val Ala Leu Gly Glu Asn Leu Glu

225

230

235

240

Ala Arg Glu Asn Met Ala Tyr Ala Ser Leu Leu Ala Gly Met Ala Phe

245

250

255

Asn Asn Ala Asn Leu Gly Tyr Val His Ala Met Ala His Gln Leu Gly

260

265

270

Gly Leu Tyr Asp Met Ala His Gly Val Ala Asn Ala Met Leu Leu Pro

275

280

285

His Val Glu Arg Tyr Asn Met Leu Ser Asn Pro Lys Lys Phe Ala Asp

290

295

300

Ile Ala Glu Phe Met Gly Glu Asn Ile Ser Gly Leu Ser Val Met Glu

305

310

315

320

Ala Ala Glu Lys Ala Ile Asn Ala Met Phe Arg Leu Ser Glu Asp Val

325

330

335

Gly Ile Pro Lys Ser Leu Lys Glu Met Gly Val Lys Gln Glu Asp Phe

340

345

350

Glu His Met Ala Glu Leu Ala Leu Leu Asp Gly Asn Ala Phe Ser Asn

355

360

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Pro Arg Lys Gly Asn Ala Lys Asp Ile Ile Asn Ile Phe Lys Ala Ala

375

380

370

Tyr

385

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<211> 35

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<220>

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<400> 9

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<210> 10

<211> 40

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40